Supplementary Materials

Establishment of MDA-MB-231 mouse xenografts, and the following administration of As^{III} and Tetra, each alone or in combination, and the preparation of tumor tissues

In vivo antitumor activity of As^{III} alone or in combination with Tetra was studied using human breast cancer nude mouse xenograft model. Five-week-old female immunodeficient BALB/c nude mice were obtained from Japan SLC, Inc. (Shizuoka, Japan) and housed at 23±1°C in a room with a constant humidity of 55±5% and a regular 12-h light/12-h dark cycle for several days. MDA-MB-231 cells (1×10^7, suspended in 0.1 ml phosphate-buffered saline (PBS)) were then injected subcutaneously into the right flank of each mouse. Tumors (visualized as a small nodule at the sites of injection) appeared approximately 5-7 days later after injection, and the mice were randomly divided into four groups (n=5) according to body weight and tumor size using SPSS 21.0 software, and given the following treatments: vehicle-control (treated with PBS); As^{III} alone (2 mg/kg/day); Tetra alone (20 mg/kg/day); As^{III} (2 mg/kg/day)+Tetra (20 mg/kg/day). The mice were administered i.p. as described above once a day for 10 weeks, respectively. The tumor size was measured every day in two perpendicular dimensions with vernier calipers, and the tumor volume (TV) (mm^3) was calculated by the formula: TV = length (mm) × $width^2$ (mm^2) × 0.5. The body weights were also measured every day and were used as an indicator of systemic toxicity of the treatment. Throughout the experiment, the mice had free access to food and water according to the National Institute of Health Guide for the Care and Use of Laboratory Animals and the Guidance for Experimental Animal Care issued by the Prime Minister's Office of Japan. At the end of the treatment, all animals were sacrificed, and the tumors were removed, photographed and weighed. Tumors were fixed in 4% paraformaldehyde (PFA) in 0.1 M phosphate buffer or frozen in liquid nitrogen for further analysis. The study was approved by the Committee of Animal Care and Welfare of Tokyo University of Pharmacy and Life Sciences (Tokyo, Japan; registration no. P17-59). Tetrandrine hydrochloride injection, and arsenious acid and sodium chloride injection were obtained from Jiangxi Yintao Pharmaceutical Co., Ltd. (Jiangxi, China) and Heilongjiang Harbin Pharmaceutical Co., Ltd. (Heilongjiang, China), respectively for the *in vivo* study.